REMARKS

Claims 23, 27-28, 32-35 and 61-69 are currently pending. Claims 1-22, 24-26, 29-31 and 36-60 are cancelled. Claims 28, 32 and 33 are amended herewith. News claim 61-63 have been added, and previously presented dependent claims 24-26 and 29-31 have been reinstated as new claims 64-69. Support for the claim amendments and new claims can be found in the claims as originally filed as well as throughout the specification. No new matter has been added.

Specification-Objections

The Examiner has objected to Figures 8 and 13 for clarity reasons. An amendment to the description of these figures as well as new drawings are entered herewith. Support for the amendments can be found in the figures as originally filed as well as in Examples 4 and 5.

The Examiner has also objected to the section numbering in Examples 6 and 7. The specification has been accordingly corrected at the Examiner's request.

Oath/Declaration

Applicants submit herewith a Supplemental Declaration in response to the request of the Examiner.

Rejection of Claims Under 35 U.S.C. §112, second paragraph

Claim 28 has been rejected "as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention." Claim 28 recites a substantially pure heparinase III comprising a polypeptide having the amino acid sequence of the mature peptide of SEQ ID NO: 2 or having conservative substitutions thereof within residues non-essential to enzymatic function, wherein at least one histidine residue selected from the group consisting of His 36, His105, His110, His139, His152, His225, His234, His241, His424, His469, and His539 has been substituted with a residue selected from the group consisting of

alanine, serine, tyrosine, threonine, and lysine. The Examiner has pointed out that the sequence of SEQ ID NO: 2 is thought to be of heparinase I rather than heparinase III.

The Applicants resubmit the sequence listing with the sequences for heparinase III.

Claim 28 is further rejected as being indefinite for having, as the Examiner asserts, two interpretations. Although the claim as written is thought to be sufficiently clear, claim 28 has been amended to provide absolute clarity. Amended claim 28 and new dependent claim 61 cover the same scope of claim 28 prior to its amendment.

Claim 28 is also rejected for reciting "conservative substitutions", which the Examiner maintains is not defined in the specification. The Examiner concedes that this term is very common in the art but that it is vague and indefinite as there is a lack of consensus on the designation of particular substitutions in this category as well as the chemical nature of particular residues.

The Applicants respectfully traverse the rejection. Firstly, the specification does provide a definition for the term "conservative substitutions". On page 12, line 22 through page 13, line 2, the definition clearly recites that a conservative substitution is one in which the substituted amino acid is of similar charge and is of similar or smaller size as the replaced residue. One of ordinary skill in the relevant art can easily determine whether or not an amino acid is of similar charge and is similar or smaller in size as another amino acid. Within the definition specific examples of conservative substitutions are also provided. Secondly, as the Examiner concedes, the term is well known and understood in the art. The art recognizes that the definition of a conservative substitution is one where an amino acid is replaced with another amino acid with similar size and chemical identity. Standard biochemistry textbooks define this term and also classify amino acids according to their properties (e.g., see Biochemistry, Geoffrey Zubay, Addison-Wesley Publishing Co., 1986 edition). The art further recognizes that whether or not a substitution is conservative depends on the impact the substitution has on the chemical and physical properties of the protein or the portion of the protein with the substitution. Therefore, the Examiner's argument that there is a lack of consensus regarding the classification of a few residues in isolated form rather than in the context of the impact on the protein does not show

that one of skill in the art could not determine whether or not use of these residues would result in a conservative change in a protein or portion thereof. One of ordinary skill in the art is able to evaluate the effect of a particular substitution on a protein or a portion thereof to determine whether or not the substitution was conservative. Therefore, based on the given definition in the specification as well as the understanding of the term and what it encompasses in the relevant art, it is believed that the term "conservative substitutions" is not vague and indefinite.

Claim 23 is also rejected as being indefinite as the Examiner asserts the claim has two interpretations. The Examiner points out that the claim could mean a composition of three independent components a heparinase III, a targeting molecule and a carrier but could also mean a composition of two independent components a fusion protein of heparinase III linked to the targeting molecule and a carrier. The Examiner concludes the specification supports the latter.

The Applicants agree that the claim encompasses both of the above interpretations. However, the Applicants assert that although the claim does encompass both interpretations, this does not mean that the claim is vague and indefinite. One of ordinary skill in the art will appreciate that the claim includes compositions where the targeting molecule is included in the composition without being linked to the other components. One of ordinary skill in the art will also appreciate that the claim encompasses compositions where the components are physically linked. The Applicants maintain that the claim is sufficiently clear as one of ordinary skill in the art is able to recognize the compositions that contain these three components regardless of whether they are linked or not.

Accordingly, Applicants respectfully request that the Examiner withdraw his rejection of claims 23 and 28 under 35 U.S.C. §112, second paragraph based on the above arguments and the amendment of claim 28.

Rejection of Claims Under 35 U.S.C. §112, first paragraph

Claims 28, 32 and 33-35 are rejected under 35 U.S.C. §112, first paragraph. Regarding, claim 28, the Examiner maintains that the specification does not reasonably enable one of ordinary skill in the art "to make and use the invention commensurate in scope with these

claims." It is asserted that although the polypeptides set forth by SEQ ID NO: 4 of US Pat. No. 5,681,733 and polypeptides with a His36Ala, His105Ala, His110Ala, His139Ala, His152Ala, His225Ala, His234Ala, His241Ala, His424Ala, His269Ala or His539Ala mutation are sufficiently enabled, a heparinase III with at least one of the histidine residues substituted with alanine, tyrosine, threonine or lysine and a heparinase III with at least one of these mutations and having any number of additional conservative substitutions are not.

The Examiner maintains that the scope of claim 28 is not commensurate with the enablement provided because of the large number of polypeptides encompassed by the claim and the only 14 representative species provided in the specification. The Examiner concludes that experimentation would be undue as no other species are provided with identifying characteristics, other than function; the effect and tolerance of changes in the sequence of heparinase III are not predictable; and it is not routine in the art to screen for the multiple modifications encompassed by these claims.

The standard for enablement is whether undue experimentation would be required for one of ordinary skill in the art to practice the claimed invention. To determine whether experimentation is undue, an analysis of the factors set forth in *In re Wands* is required. *In re Wands*, 858 F.2d 731, 8 USPQ2d 1400 (Fed. Cir. 1988). The Examiner has not analyzed each one of them in order to reach the present conclusion. The Applicants assert that based on the arguments presented by the Examiner, experimentation is not undue.

The Applicants assert that the number of species provided is sufficient, and that in view of the guidance and teachings found in the specification the claimed substitutions are predictable and it is routine in the art to screen for multiple modifications represented by the claims. Claim 28 provides a set of residues that can be modified to produce molecules with altered activity. The recitation of these residues in combination with the teachings of the disclosure and extensive working examples provides adequate guidance to one of skill in the art to produce a modified heparinase III and to test for a desired activity. The specification, working examples and level of skill in the art provide sufficient guidance for one of ordinary skill to identify, make and test the

modified heparinase III molecules and, therefore, use the molecules as stated in the claim, with only routine experimentation.

One of skill in the art is able to identify heparinase III molecules useful according to the claimed methods and to make modified heparinase III molecules with the teachings provided by Applicants' specification and the high level of knowledge in the art. For example, the specification provides the nucleic acid sequence of heparinase III, which was known in the art, with the recitation of references Su et al., 1996 and Godavarti et al., 1996; US Patent Nos. 5,919,693 and 5,681,733 and Accession Number I71365 on page 9 of the specification.

One of skill is sufficiently enabled to produce modified heparinase III molecules with standard techniques known in the art and the guidance provided in Applicants' specification through the identification of the residues that are important to the function of the enzyme. An adequate and extensive description of the histidine residues that can be modified in heparinase III are provided. Through the description of these residues that are involved in the function of the enzyme, the specification provides the structure of heparinase III in relation to its function towards heparan sulfate. The claims specifically recite these residues. In addition, the specification provides a representative number of species that demonstrate the various mutations which alter the enzymatic activity of the enzyme. One of skill would be able to produce modified heparinase III molecules with altered activity due to the recitation of the specific residues to be substituted in the amended claims as well as the description, working examples and lists of substitutions provided in the specification.

One of ordinary skill in the art is also sufficiently enabled to further modify the heparinase III polypeptides with conservative substitutions within residues non-essential to enzymatic function. Conservative substitutions as argued above are well known in the art. Additionally, the amount of experimentation that is required to make a modified heparinase III with conservative substitutions and to test for the desired activity is routine in the art in view of the teachings in the specification.

Techniques for determining modified product profiles and/or k_{cat} values are well known in the art, and examples of these techniques are also given in the specification (e.g. pages 14 and

15-17). For example, a method of using mass spectrometry and capillary electrophoresis is described for determining the product profile of a heparinase. Other methods provided for determining product profiles rely on viscosity, total UV absorbance or mass spectrometry or capillary electrophoresis alone. Enzymatic activity assays for determining the k_{cat} value of a heparinase enzyme are also described in the specification. In view of this, one of skill would be required to perform only routine experimentation to screen modified heparinase III enzymes having the structures recited in the claims.

Because of Applicants' description of mutant heparinase III enzymes as well as the residues that can be modified, the disclosure provides a clear finite set of possible modifications of heparinase III. Applicants further maintain that the specification also provides sufficient guidance for testing the modified heparinase III enzymes produced to select those for a specific property. The specification provides sufficient guidance to one of ordinary skill in the art to make and use the invention commensurate in scope with these claims.

It is further asserted that the specification does not provide enablement for any modified heparinase III having a modified product profile (with any substrate) or a k_{cat} value (with heparan sulfate as the substrate), that is at least 10% different than native heparinase III. Claims 32 and 33 have been amended to recite a modified heparinase III having specific amino acid residue substitutions that result in the modified heparinase III having a modified product profile and/or modified k_{cat} for use in the claimed methods (as in claim 28). The modified heparinase III molecules may also have additional conservative substitutions of residues not necessary for the enzyme's function. Thus, the claims now recite structural limitations which have been identified according to the invention to produce the functional properties of the claimed enzymes. Accordingly, because of the arguments presented above for claim 28, the Applicants maintain that amended claims 32 and 33 should now also be in a condition for allowance.

The Examiner has further rejected claims 28 and 32-35 as not being described in the specification in such a way as to demonstrate to one of skill in the art that the Applicants were in possession of the claimed invention.

The Applicants respectfully disagree. The disclosure of the amino acid residues that play a role in the activity of heparinase III along with the number of described species is sufficient to demonstrate Applicants were in possession of the claimed invention.

In the specification, several representative species are disclosed which include modified heparinase III molecules with mutations at 13 histidine residues within heparinase III. These modified heparinase III enzymes exhibit differential activity toward heparan sulfate. For example, mutation of His 295 or His 510 results in a modified heparinase III enzyme with no activity toward heparan sulfate. The His 110 and His 241 mutants are less active towards heparan sulfate, while the His 139 mutant has increased activity. The His 225 mutant displays nearly the same enzymatic activity as the native enzyme.

Based on the foregoing, the specification provides an adequate description of the invention to demonstrate that Applicants had possession of the claimed invention as well as adequate information about the structure of heparinase III as it relates to the function of the enzyme.

Accordingly, Applicants respectfully request that the Examiner withdraw his rejection of claims 28, 32 and 33-35 under 35 U.S.C. §112, first paragraph based on the above arguments and the amendment of claims 32 and 33.

Rejection of Claims Under 35 U.S.C. §102(b)

Claim 23 is rejected under 35 U.S.C. §102(b) as being anticipated by Bennett et al., 1997. Bennett et al. teaches the targeting of heparinase III through fusion proteins derived from enzymes and binding domains from antibodies, growth factors or adhesion molecules.

Applicants respectfully traverse the rejection of claim 23. For a reference to be anticipatory, the reference must recite all of the limitations of a claim. In this case, the claim is to a composition of a heparinase III or a therapeutic HLGAG fragment in an effective amount for preventing metastasis of a tumor cell and a targeting molecule for targeting the heparinase III to the tumor, in a pharmaceutically acceptable carrier. However, Bennett et al. does not teach an effective amount of a heparinase III to prevent metastasis of a tumor cell nor a targeting

molecule for targeting heparinase III to a tumor cell. Bennett et al. merely teaches a heparinase III that is targeted to activated endothelial cells to reduce an inflammatory response. Therefore, because Bennett et al. fails to teach the above limitations present in claim 23, Bennett et al. does not anticipate this claim.

Claims 32 and 33 are also rejected under 35 U.S.C. §102(b) as being anticipated by Lohse et al., 1992. Lohse et al. teaches a heparinase II, which uses heparan sulfate as a substrate, as has a higher K_m and lower V_{max} when compared to heparinase III. Because of the different kinetic properties it is asserted that the action of heparinase II on heparan sulfate would have an altered product profile and a k_{cat} that is at least 10% different than that of native heparinase III. As claims 32 and 33 fail to recite any structural limitations for the modified heparinase III, heparinase II would read on the modified heparinase III molecules of these claims.

The Applicants respectfully disagree with the Examiner's conclusion regarding these claims as the claims are intended to encompass modified heparinase III and not native heparinases. However, as the claims have been amended as described above, the need to further argue this rejection has been obviated.

Accordingly, Applicants respectfully request that the Examiner withdraw his rejection of claims 23, 32 and 33 under 35 U.S.C. §102(b) based on the above arguments and the amendment of claims 32 and 33.

Rejection of Claims Under 35 U.S.C. §103

Claim 28 has been rejected under 35 U.S.C. §103(a) as being unpatentable over Su et al, 1997 US Pat. No. 5,681,733 in view of Shriver et al., 1998 as Su et al. provides the protein sequence of heparinase III and Shriver teaches that specific histidine residues of heparinase II are important for enzymatic activity against its substrates heparin and heparan sulfate. The Examiner concludes that one of ordinary skill in the art would predict that heparinase residues are important for the activity of heparinase III, and therefore, be motivated to the methods of Shriver to prepare the mutants to determine which residues regulate the activity of heparinase III.

The Examiner further states that the expectation of success is high as site-directed mutagenesis is common in the art. The Applicants respectfully disagree with this assessment.

The Shriver et al. teachings are directed to heparinase II, a completely different protein than heparinase III with different structural and chemical properties. Heparinase II and heparinase III have different binding affinities and different substrate specificities. It would, therefore, not have been obvious to one of ordinary skill in the art that the same residues that impact heparinase II's function would necessarily be the same as the residues responsible for the activity of heparinase III.

Further, the two references do not even teach or suggest the specific histidine residues that can be substituted nor the amino acids that can be used for the substitution all of which are recited in the claims. Claim 28 recites 11 specific histidine residues that can be substituted to produce modified heparinase III molecules. The claim further recites that these residues are substituted with alanine, serine, tyrosine, threonine, or lysine. The Examiner has not provided any evidence that these claim limitations are taught in the two references. The Examiner also provides no evidence that one of ordinary skill in the art would have a reasonable expectation of success in obtaining the claimed invention. The Examiner points out that because the level of skill is high and that the use of site-directed mutagenesis in the art is common there is a reasonable expectation of success. However, this expectation speaks only to the ability to produce mutants but not to obtaining the claimed invention. The Examiner has provided no evidence that one of skill in the art could predict the claimed invention (i.e., the specific histidine residues recited that are substituted with alanine, serine, tyrosine, threonine, or lysine).

Finally, the Examiner also provides no evidence that the art suggests the desirability to combine these two references other than a general desire to determine whether or not histidine residues regulate the activity of heparinase III. There is no evidence that points to the desirability of combining the two references other than this general notion. At best, the combination of the above references provides the motivation to try to determine the importance of the histidine residues in heparinase III. However, a motivation to try is not sufficient to satisfy a *prima facie* case of obviousness.

Dependent claims 34 and 35 are also rejected as being unpatentable under 35 U.S.C. §103(a). However, as the rejection of these claims are premised on the obviousness of the Su et al. and Shriver et al. references (in combination with additional references) and the Applicants maintain that the *prima facie* case of obviousness with the Su et al. and Shriver et al. references has not been established, the Applicants maintain that the arguments presented above are also sufficient to overcome the rejection of these dependent claims.

Accordingly, Applicants respectfully request that the Examiner withdraw his rejection of claims 28, 34 and 35 under 35 U.S.C. §§103(a) based on the above arguments.

Rejection of Claims Under 35 U.S.C. §101, double patenting

Claims 32 and 33 are rejected under 35 U.S.C. §101 as being unpatentable over claim 1 of US Pat. No. 5,389,539 which is a claim to a heparinase II having a molecular weight of 84,100, a pH optimum of 8.9-9.1 and cleaves heparin and heparan sulfate. The rejection is based on the premise that the heparinase II is a modified heparinase III molecule with heparan sulfate as a substrate. The Examiner concludes that as claim 32 and 33 are to a modified heparinase III with modified product profile or a k_{cat} value, respectively, that is at least 10% different than a native heparinase III and do not recite any structural limitations, the heparinase II of the '539 patent reads on the modified heparinase III molecules of these claims.

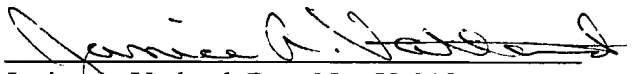
Although the Applicants respectfully disagree with the Examiner's conclusion regarding these claims, as the claims are intended to encompass modified heparinase III and not native heparinases, and as the claims have been amended the need to further argue this rejection has been obviated.

Accordingly, Applicants respectfully request that the Examiner withdraw his rejection of claims 32 and 33 under 35 U.S.C. §101 based on the above arguments and the amendment of claims 32 and 33.

Summary

It is believed that all of the pending claims are in condition for allowance. If the Examiner has any questions or comments, he is encouraged to contact Applicants' representative at the number listed below.

Respectfully submitted,



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